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Effects of Simulated Low-Altitude Aircraft Overflights on White-Leghorn Broilers and Laying Hens

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Interim Report for the Period October 1997 to June 1999

Human Effectiveness Directorate
Warfighter Interface Division
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NOTICES

TECHNICAL REVIEW AND APPROVAL

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The experiments reported herein were conducted according to the "Guide for the Care and Use of Laboratory Animals," Institute of Laboratory Animal Resources, National Research Council.

This report has been reviewed by the Office of Public Affairs (PA) and is releasable to the National Technical Information Service (NTIS). At NTIS, it will be available to the general public.

This technical report has been reviewed and is approved for publication.

FOR THE COMMANDER

//Signed//

LEE D. SHIBLEY, Lt Col, USAF
Deputy Chief, Warfighter Interface Division
Air Force Research Laboratory

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14. ABSTRACT

Low-level overflights and sonic booms have been suspected of having serious effects on domestic fowl. Some of these effects have not been produced experimentally, despite repeated attempts. Panic effects such as piling and crowding have been the focus of several recent studies, but they did not determine the threshold for this response. The present program included experiments on two cohorts of naïve broilers, young (3 weeks) and older (8 weeks), and Cobb white-leghorn laying hens. Poult weight gain and carcass quality were also examined in birds exposed during this study. The program collected heart rate measurements of young broilers during and after exposure to a series of simulated overflights in an effort to obtain a physiological estimator of broiler response to overflight stimuli. The broilers were exposed to simulated overflights that varied in sound exposure level, onset time, duration and interval between exposures. Such measurements had not been made in poultry prior to these experiments. Hens and broilers did not experience excess mortality or changes in weight as a result of exposure. Egg production was not affected by simulated overflights, nor was egg quality.

15. SUBJECT TERMS

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PREFACE

The work reported herein was conducted by Hubbs-Sea World Research Institute, San Diego CA, through subcontract from Veridian Engineering, Dayton OH, under Air Force contract F41624-95-C-6014, program element 62202F, work unit 71841611. The program was managed in the Battlespace Acoustics Branch, Human Effectiveness Directorate, Air Force Research Laboratory, Wright-Patterson AFB OH. Robert A. Lee, branch chief, was the technical monitor for the effort.

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I. FOREWORD

This report was prepared by Dr. Ann E. Bowles and the staff of Hubbs-Sea World Research Institute under contract to Veridian, in fulfillment of purchase orders P3835, P3829, K3202. The research was supported by the USAF, Air Combat Command, Brooks Air Force Base, and the USAF Armstrong Laboratories, Noise Effects Branch, at Wright-Patterson Air Force Base (Contract No. F41624-95-C-6014 DO-C9). The project was managed by Captain Michael Carter (USAF, Armstrong Laboratory, Noise Effects Branch [AL/OEBN]) and Mr. Alva Karl (Veridian).

All procedures were approved by Institutional Animal Care and Use Committees at the U.S. Air Force and Hubbs-Sea World Research Institute.

Animal resources and technical support were provided by the Department of Animal Science at the Oklahoma State University, Stillwater, Oklahoma, under the direction of Dr. Robert Teeter.

II. ACKNOWLEDGMENTS

Many people assisted us in the conduct of this study. The staff and students of the Oklahoma State University Department of Animal Science Poultry Science Research Farm provided invaluable assistance under the direction of Mr. Farzad Deyhim. Dr. VanHooser kindly implanted heart rate monitors in young broilers. Lanelle Caldwell did the data entry and helped us interpret formats. Captain Michael Carter gave us his time and energy when we most needed it, assisting in the conduct of playback studies on laying hens and acting as a go-between in Oklahoma during analysis. Mr. Bill Russell with the US Army/CERL kindly provided us with the four Larson-Davis community noise monitors needed for this study. Rick Rushton at MiniMitter provided us with technical information on the PhysioTel heart rate monitoring system that was invaluable.

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III. TERMS AND ABBREVIATIONS

Unless otherwise specified, all acoustical terms are drawn from Harris (1991) and ANSI S1.1-1994.

A/D	analog-to-digital converter
AEP	auditory-evoked potential
ANSS	Aircraft Noise Simulation System
ASEL	A-weighted sound exposure level
BPM	heart beats per minute
CSEL	C-weighted sound exposure level
cm	centimeter (metric abbreviation)
D/A	digital-to-analog converter
DAT	digital audio tape
ECG	electrocardiogram
FFT	fast Fourier transform
HSWRI	Hubbs-Sea World Research Institute
Hz	Hertz, cycles per second
IACUC	Institutional Animal Care and Use Committee
L_{max}	maximum, fast, A-weighted sound pressure level
ms	milliseconds (metric abbreviation)
Pa	Pascals
peak	largest absolute value of sound pressure
psf	per square foot
s	seconds (metric abbreviation)
SEL	sound exposure level
SNR	signal-to-noise ratio
sound level	fast, A-weighted sound pressure level.

SPL	sound pressure level.
USAF	U.S. Air Force
wks	Weeks

IV. SUMMARY

Based on veterinarian accounts of piling and crowding incidents, domestic chickens are capable of panicking in response to aircraft overflights (*e.g.*, Milligan *et al.* 1983); however, experimental efforts have failed consistently to elicit this response. The reasons for the difference between experimental and industrial conditions are poorly-understood. In addition, changes in behavior and physiology after exposure have not been described. In 1997, experiments were conducted under USAF funding to examine the effects of simulated low-altitude overflights on behaviors and productivity of white leghorn broilers and Cobb-Vantress laying pullets. Given the difficulty of arousing piling and crowding, ethical concerns, and the expense of conducting research in an industrial setting with flocks numbering thousands or tens of thousands, the program was designed not to cause damaging incidents, but to obtain data necessary to improve models of the probability of such incidents.

Experiments were conducted at the Poultry Science Research Laboratory at Oklahoma State University in Stillwater. Dr. Robert Teeter with the Department of Animal Science provided facilities and technical assistance. Birds were exposed to simulated aircraft overflights designed to obtain a detailed model of the relationship between exposure level and bird behavior. In addition, measurements were made to determine whether there were longer-term changes in behavior and productivity were associated with exposures. The birds were housed in small groups (up to 60 birds) in separate pens in two poultry houses at OSU. Level of exposure varied with pen distance from the projecting system. The first set of trials used older broilers (8-week-old white leghorns) because larger birds were thought to be more likely to pile and crowd dangerously during naive flight responses. However, when these birds could not be stimulated to flight at any sound level, further trials were conducted with younger birds (3-week-old white leghorn broilers). In addition, effects on behavior and productivity of Cobb-Vantress laying hens were measured.

Stimuli were delivered with a USAF Sound Simulation System mounted overhead at one end of the test barn. Attenuation within the barn was measured with four LD 820 community noise monitors provided by the Noise Effects Branch, Armstrong Laboratory, at Wright-Patterson Air Force Base. Pens were broken in to 4 sets of exposure categories corresponding to 4 sections of the barns (1: high, up to 7.5 dB attenuation; 2: medium up to 10 dB; 3:low up to 15 dB; and 4: ambient ~ 20 dB). Audibility of the experimental exposures in the 4th section depended on levels of fan noise and vocalizations from the chickens.

The following experiments were conducted:

- 72 pens of 8-week-old broilers (1884 birds) were exposed to 4-6 simulated jet overflights per day with levels up to 115 dB ASEL. Behavior, food and water consumption, and changes in body weight were measured.
- 72 pens of 3-week-old broilers (4388 birds) were exposed to 4 simulated jet overflights per day with levels up to 115 dB ASEL. Behavior, food and water consumption, and changes in body weight were measured.
- 6 broilers were fitted with implanted heart rate monitors and exposed to 14 simulated overflights per day for 2 weeks at varying levels.
- 56 pens of laying hens were exposed to two series of simulated overflights separated by four days of blank trials over a 2 week period (hens 48-50 weeks of age). Hen behavior, weight, food consumption, egg production, and egg quality were monitored.

Initial jet simulations at the highest levels stimulated 8-week old broilers to stand, aggregate, and search for the source of the sound (orienting). Although a few individuals ran briefly, none of the groups piled and crowded. After the 8-week-old broiler experiments, HSWRI and OSU staff considered possible explanations for failure to panic - broilers (1) were past the age of maximal responsiveness, (2) were accustomed to human-made disturbances, or (3) were in groups that were below some 'critical mass' needed for dangerous piling and crowding. Of these, the first was deemed highly probable, so a second series of experiments with smaller birds was planned.

With repeated exposure, birds ceased to stand or aggregate. By the last day of experiments, broilers looked up or simply remained still during the stimuli. However, they exhibited a highly stereotypical sequence of events after the exposures, regardless of their initial reaction. At 1-2 min post-exposure, they relaxed. At 2-4 min, they preened more, drank more and fed more than during the pre-stimulus period. From 4-8 minutes, they were more active and engaged in more aggressive encounters. After this, they returned to baseline activity levels.

This sequence of events was apparently an innate response to disturbance. Entry of humans into the pen to care for the broilers produced a similar sequence of events in all the birds tested.

During the 8-week-old broiler experiments, incidents of picking were quantified by collecting counts of birds with comb wounds or blood on their feathers from each pen twice per day (14 observations/pen). These incidents were seen most often in the sections of the barn where human activity was greatest, but also correlated with noise exposure. In the low exposure section of the barn, 48 birds were seen with blood spots (section 3); 62 were seen in the moderate exposure section (2) and 98 were seen in the high exposure section (1). Human activity was highest in section 4, which also experienced elevated ambient noise levels from the ventilation system; 91 birds were observed with blood marks in this section. One bird died due to picking in the high-exposure section (1). Although human activity confounded the results of these measurements, the data led to an important conclusion – picking results directly from an increase in activity that brings birds into contact, resulting in higher levels of aggression.

Laying hens and 3-week-old broilers were rarely aggressive to one another and did not cause picking injuries during the course of these experiments.

No detectable differences in growth rate or mortality were observed among exposure conditions in 8-week old broilers. These broilers experienced high mortality; an average of 14% of the birds in the test died of various causes. The birds were just past the age of slaughter for a commercial operation; at this age, growth is very rapid and crowding becomes an important factor. No significant differences in mortality were found among different exposure groups, however (ANOVA, $p \gg 0.05$; the highest mortality was found in the low-exposure section). Broilers grew by an average of 0.58 lb during the experiments, but there were no significant differences among the sections in weight gain (ANOVA, $p \gg 0.05$), despite differences in initial and final weight.

No detectable differences in growth rate or mortality were observed among exposure conditions in 3-week old broilers.

Hens had a known laying history before the start of experiments. They were exposed to 4 overflights per day for 5 days, blank trials for 4 days, and overflight exposures for another 4 days. No difference was found between observed and expected laying rate during all exposure days combined or between exposed days and days with blank trials.

Because a previous study had found blood spots in eggs of hens exposed to continuous noise, there was concern that exposure to aircraft noise might affect eggs. On one day before exposure and one day after the first 5-day block of exposures (which would have been most likely to alarm hens) eggs were candled to determine whether they contained blood spots or tissue spots. Blood spots and tissue spots were never common. In the pre-exposure candling, 0.4% had blood spots and 0.5% had tissue spots (of 790 eggs). There was no significant difference post-exposure (0.1% had blood spots and 0.1% had tissue spots out of 740 eggs).

In summary:

- Hens and broilers did not experience excess mortality or changes in weight as a result of exposure.

- Hens and broilers exhibited a highly-stereotypical sequence of behaviors after exposure, including increased eating, drinking, and agonistic (=aggressive) interactions (these last were observed in 8-week old broilers only).

- No piling and crowding incidents were observed in the small groups that were tested (pens had up to 60 individuals).

- An increase in picking was observed in older (8-week-old) broilers that was not observed in younger broilers or hens. The number of birds encountered with marks of picking correlated with levels of noise exposure, but also human activity in the barn.

- Egg production was not affected by simulated overflights. No increases in eggs with double yolks, blood spots or tissue spots were observed.

Physiological measurements (heart rate and metabolic rate) and effects on feed and water consumption have not been analyzed fully as yet.

The only likely effect of simulated overflights on broilers observed during this study was an increase in picking by older broilers. This effect was also observed in an earlier study of turkey poults (Bradley *et al.* 1990). Picking reduces carcass quality and bird welfare. The influence of simulated overflights on aggression could not be quantified because human activity associated with husbandry activities and experiments confounded the effect. The degree of loss was small, and was not observed in younger broilers and laying hens. Because the older broilers were beyond the normal age of slaughter, the effect may not appear in commercial operations. The important conclusion to be drawn is not that picking represents a large economic risk to commercially-raised broilers, but that in cohorts of poultry likely to become aggressive repeated disturbances can give rise to incidents of picking and these incidents will be seen most often at high levels of exposure (>90 dB ASEL).

The result makes poultry responses to disturbance clearer. Arousal and defensiveness stimulated by aircraft noise cause birds to rise and congregate defensively. Once aroused, they eat, and drink, behaviors that may bring them into even closer proximity. Agonism (threats and pecking) result. The effect was detectable as a result of accumulated exposure - the impact of single incidents was small enough that it would have been difficult to detect.

Failure to pile and crowd remains unexplained. Aggregation, a precursor to this behavior, was observed during the first few exposures in the most heavily exposed groups. When analysis is complete, aggregation (3 or more birds moving within 1 body length) will be treated as an indicator of response in the development of dose-response models. Failure to pile and crowd was not a result of inadequacy of the simulations - in experiments on turkey poults (Bradley *et al.* 1990) dangerous piling and crowding was aroused easily by simulated overflights. Therefore, it seems unlikely that susceptibility to panic in chickens is determined by age class, sex, or small differences in genetic makeup (hens were from a different strain than broilers). It seems most likely that chickens simply do not have tendency to panic as strong as that of turkeys. This is consistent with the much longer history of domestication for this species.

V. INTRODUCTION

Aircraft noise, in the form of low-level overflights and sonic booms, has been suspected of having many serious effects on domestic fowl, including 1) causing egg breakage and reduced hatchability, 2) lowering productivity or the productive lifetime of laying hens, 3) inducing panics, which can result in crowding, piling and smothering; and 4) reducing growth rate. Some of these effects, particularly egg breakage and reduced hatchability, have not been produced experimentally, despite repeated attempts (Stadelman, 1958b, Heinemann and LeBrocq, 1965, Cottereau, 1972, Cogger and Zegarra 1981, Von Rhein 1983, Bowles *et al.*, 1991, Bowles *et al.*, 1994). Other effects, including deaths and injuries due to panic crowding and piling, are likely to be important in areas where birds are unfamiliar with aircraft noise (naïve). Such losses are well substantiated (Milligan *et al.* 1983, Bradley *et al.* 1990, unpublished data in the USAF Claims Files).

Because panic piling and crowding are likely to have a significant economic impact on poultry growers and because they can be induced experimentally, panic effects have been the focus of several recent studies. Bradley *et al.* (1990) and Von Rhein (1983) succeeded in stimulating piling and crowding responses (with simulated and real aircraft overflights, respectively), but did not determine the threshold for this response. In both of these studies, broilers and turkey poults were exposed at 2-3 weeks of age.

The present program included experiments on two cohorts of naïve broilers, young (3 week old) and older (8 week old). Playback experiments with these broilers were primarily designed to improve an existing dose-response model for domestic fowl. However, because a previous USAF study found an effect on carcass quality of another domestic species, the Nicholas strain turkey (Bradley *et al.*, 1990), poult weight gain and carcass quality were examined in birds exposed during this study as well.

Mortalities were not expected as a result of the experimental protocol. Piling and crowding are species-typical antipredator behaviors in flocking birds and are exhibited in response to many natural and human-made stimuli. Poultry frequently pile and crowd without causing deaths. A number of environmental factors (weather, housing, *etc.*) contribute to actual mortalities. For example, Milligan *et al.* (1983) cited cases in which large numbers of birds were lost as a result of unusual weather (a heat wave) or housing conditions. In another study (Von Rhein 1983), water deprivation contributed to losses. Experiments exposing birds to these conditions would be difficult to justify within the guidelines of HSWRI and USAF Institutional Animal Care and Use Committees (IACUC), nor could all the possible combinations of factors be tested. Therefore, the present studies of broilers (3 wks and 8 wks) and laying hens were designed to measure the threshold for crowding responses in naïve broilers and laying hens. The actual rate of losses once birds panic must be estimated using grower incident reports and previous claims against the USAF.

Changes in egg productivity and egg quality as a result of exposure to transients such as aircraft overflights have been cited in claims against the U.S. Air Force (USAF Claims files), but have not been demonstrated in experimental studies of laying chickens (Cogger and Zegarra 1981, Stadelman 1958a, Von Rhein 1983) or in well-documented clinical accounts. Effects on broodiness have been found in studies of other species (the domestic turkey; Jeannotout and Adams 1961), and effects on laying have been found in chickens exposed to continuous or nearly-continuous poultry-house noise at high levels (average levels > 80 dB SPL; Belanovskii and Omel'yanenko 1982). Egg quality has also been affected by high levels of continuous exposure (Kagan and Ellis 1974). Unfortunately, these effects either do not occur after transients or with such low incidence that they have proved elusive to document. It is possible that previous efforts to demonstrate effects did not present hens with the appropriate stimuli or that the measurement techniques used were inadequate to detect subtle differences in numbers of eggs laid and egg quality. As a result, the present program included experiments on Cobb white-leghorn laying hens.

In previous experiments with poultry, overt behavioral responses habituated quickly, making it difficult to measure the relative importance of overflight parameters such as level and duration. Since these parameters can strongly affect responsiveness of laboratory animals, it was desirable to find a way to measure responses from the same individual birds repeatedly. This program collected heart rate measurements of young (3-week old) broilers during and after exposure to a series of simulated overflights in an effort to obtain a physiological estimator of broiler response to overflight stimuli. The broilers were exposed to simulated overflights that varied in sound exposure level, onset time, duration, and interval between exposures. Such measurements had not been made in poultry previous to these experiments due to the difficulty of obtaining heart rate measurements from unrestrained birds.

VI. METHODS

ACOUSTIC MONITORING AND SOUND PLAYBACK

Four U.S. Army Larson-Davis 820 (LD 820) community noise monitors and two USAF Aircraft Sound Simulation Systems (USAF/ANSS) were made available to the program by the U.S. Army Bio-Acoustics Division, Aberdeen Proving Grounds and the Armstrong Laboratory, Wright-Patterson Air Force Base. These were deployed in the two test barns used for the study (a 72-pen house used for broiler experiments and a 60-pen house used for laying hen experiments) and in the laboratory used for physiological measurements.

The two USAF/ANSS were used to generate 6 simulated overflights with peak levels directly under the speakers exceeding 110 dB (103 dB ASEL; Table I). They were mounted facing downward at one end of the barn, with levels set before the start of experiments to achieve a peak of approximately 110 dB ASEL for the most intense overflights. These settings were established in the absence of chickens; levels were measured by an LD 820 mounted 2 m below the speakers and 1.2 m above the floor. Sound propagated towards the other end of the barn, which was equipped with three large blowers to draw fresh air through the barn (Figure 1). The absolute level and signal to noise ratio of playbacks declined across the barn; at the far end, levels often did not exceed ambient. The level in each pen was determined as follows. White noise was projected from the speakers in the absence of chickens and levels in the center of each pen were measured using an LD 820. The difference between level measured directly under the speaker and level in the pen was the attenuation for each pen. Barns were broken in to 4 sections corresponding to 4 exposure categories (1: high, up to 7.5 dB attenuation; 2: medium up to 10 dB; 3: medium 15 dB; and 4: ambient ~

Table I. Features of playback stimuli.

Stimulus	Peak Level (dB)	ASEL (dB)	Duration (s)	Onset Time
1	125.1	108.9	21.96	High
2	119.5	112.3	30.28	High
3	122.9	107.5	32.93	High
4	120.5	113.4	19.31	Low
5	112.8	103.4	40.31	Low
6	126.1	114.9	24.68	Low

20 dB). Audibility of the experimental exposures in the 4th section depended on levels of fan noise and vocalizations from the chickens.

The 72-pen barn was 165 ft in length, with attenuation ranging from +1 dB under the speakers to ~ -20 dB at the opposite end of the barn. The 60-pen barn was 214 ft in length, with attenuation ranging from +1 dB under the speakers to -25 dB at the far end (Figure 1). Four LD 820 meters were deployed throughout the experiments, one in the middle of each section, to quantify ambient noise and level of experimental overflights.

Real-time recordings were also collected to obtain detailed information on noise in the barns, including noises produced by equipment and the birds themselves.

The sound playback stimuli were pre-recorded jet overflights of varying level, duration, and onset time (Table I). It was recognized that some acoustic components of an overflight could not be simulated, particularly low-frequency components (<~50 Hz), the two-dimensional sound field, and unpredictability. Every effort was made to compensate for these differences. Multiple stimuli were used to slow habituation. Each overflight was phased between the two sets of USAF/ANSS speakers to simulate the aural illusion of aircraft movement as much as possible. Intervals between exposures and the order of exposure was also varied to make onset of the experiment unpredictable.

Some differences could not be eliminated, particularly differences in the sound field. If a real aircraft had overflown the farm, all pens would have received roughly the same sound level, while levels varied greatly from one end of the barn to the other during simulations. This property of the playback protocol was used deliberately to obtain samples of flock reactions to a wide range of levels, but it eliminated reactions that might have been triggered if all individuals had been exposed similarly.

The simulated overflights were also more predictable than a real aircraft because few simulated overflights (6 in total) were delivered and the perceived behavior of the aircraft did not vary (it was always heard to travel the same route). In contrast, a real aircraft has an infinite number of idiosyncratic variations, produced by differences in flight attitude, engine power setting, and so on. However, simulations of the type used have successfully aroused panic responses in turkey poults (Bradley *et al.* 1990) and were convincing facsimiles from the perspective of human listeners.

Calibrations were completed at the end of the trial series in each barn by projecting white noise from the USAF/ANSS at a known level (84.5 and 88.7 dB SPL in the 72-pen and 60-pen

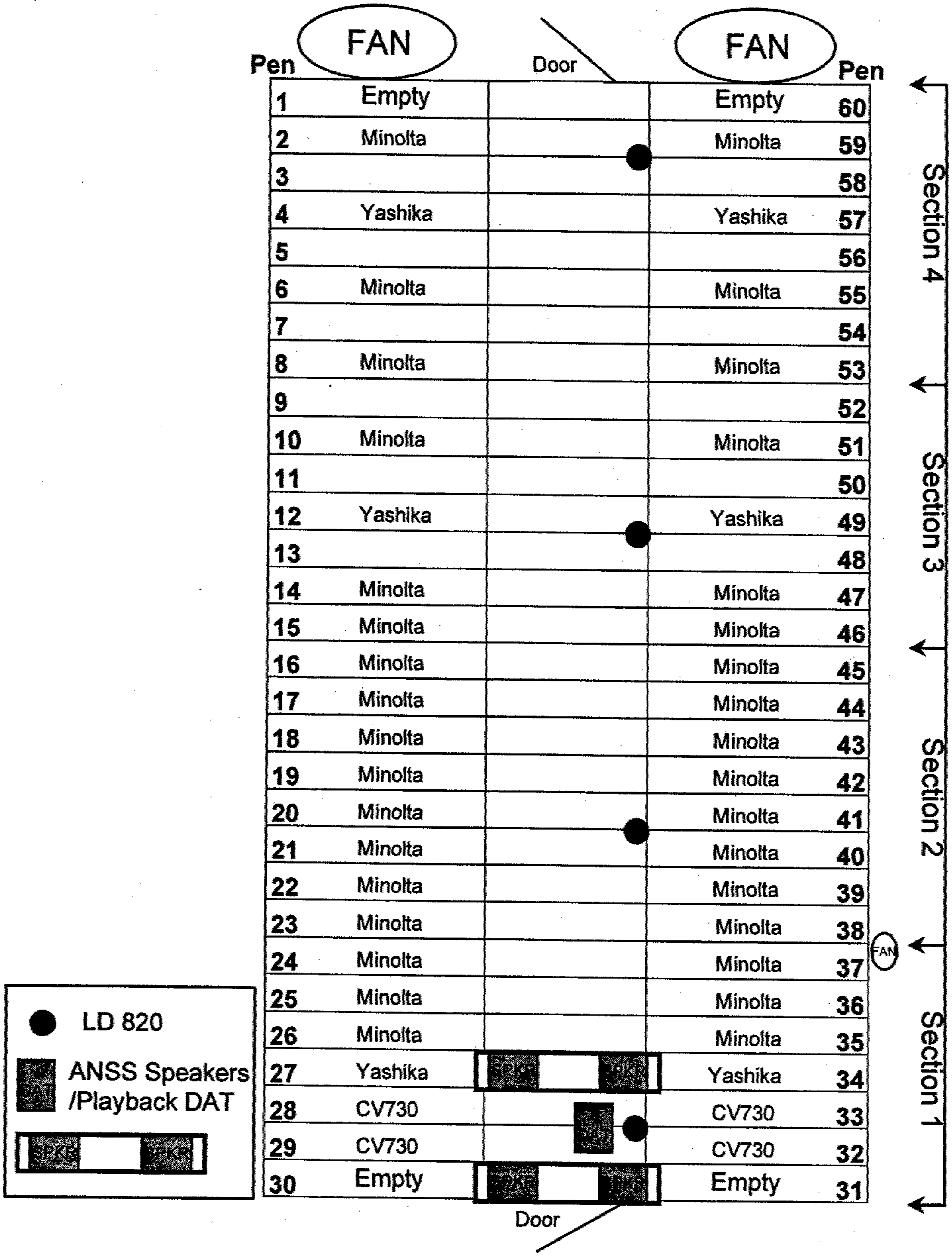

Figure 1. Layout of 60-pen house. Locations of ANSS System, LD 820 monitors, and overhead cameras (CV730 video, Yashika and Minolta still cameras) are shown.

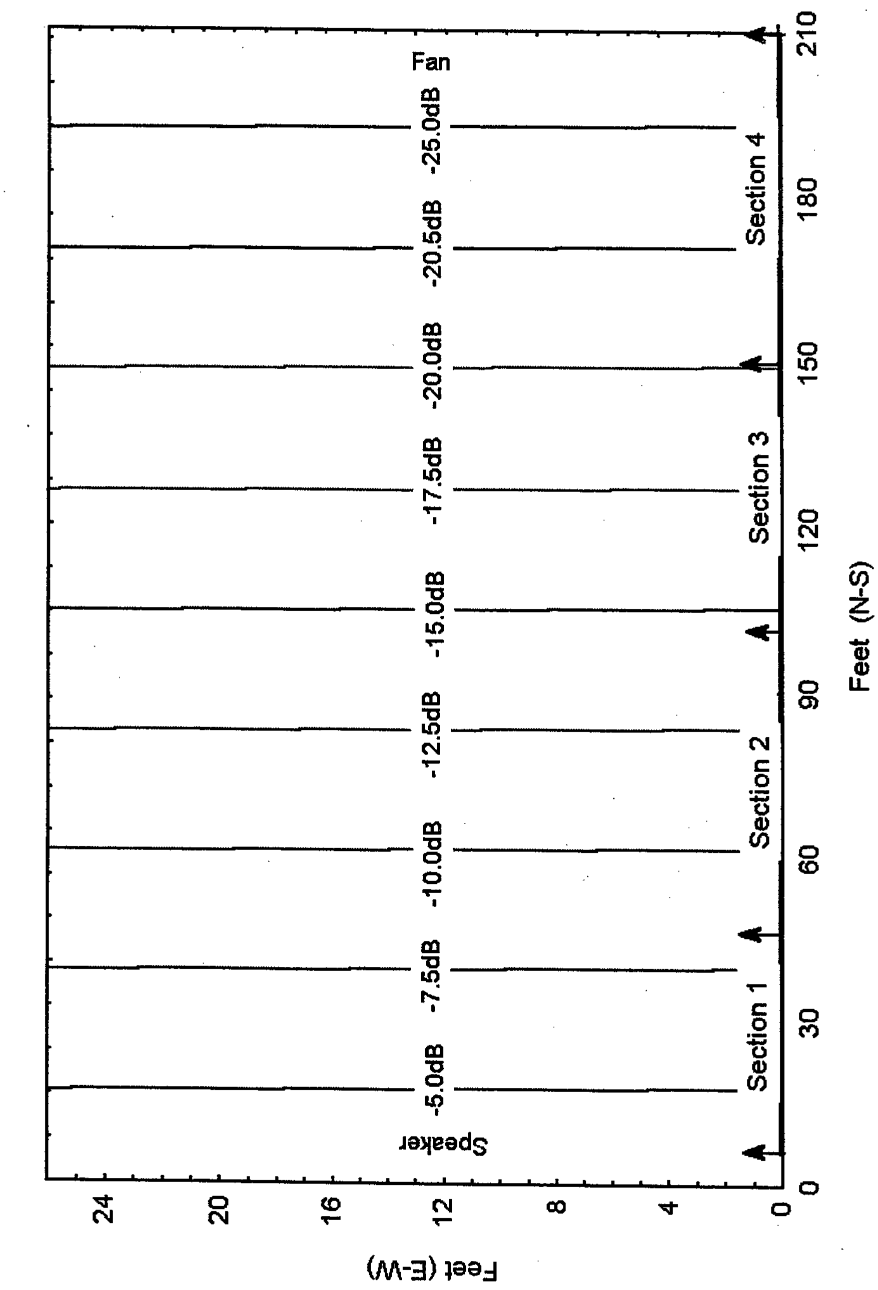

Figure 2. Sound calibration map for 60-pen poultry house. The white noise used to obtain these attenuation measurements had an A-weighted SPL of 88.7 dB at 1 m from the speakers.

houses, respectively). The level of this noise was measured in the center of every pen and a calibration map was produced for both barns (Figure 2).

MEASUREMENTS OF BROILER AND HEN RESPONSES

All birds used in these experiments were made available after a series of experiments on feed formulas/intake at the Poultry Science Research Farm. Both younger (3-week old) and older (8-week old) broilers were tested in a poultry house with 72 pens. Laying hens were made available immediately after reaching the peak of laying efficiency (48 weeks), and were tested in a house with 60 pens.

Both the 72-pen and 60-pen houses were broken into 4 sections corresponding roughly to four different exposure regimes (high, high-medium, medium, low). Eight pens in the section with highest exposure levels were monitored using time-lapse video. Other pens throughout the barn were be monitored using still cameras (Figure 1) that were triggered simultaneously using a pneumatic triggering system. The triggering system made an audible but not intense clicking noise during activation. In order to insure that these sounds did not affect broiler behavior, "blank" trials with a blank tape were conducted before the start of experiments and about once per day during the experimental series. As far as observers on-site could determine, the chickens were well-habituated to the experimental procedures, including the clicking of the pneumatic trigger, equipment manipulation, and the presence of observers, for two reasons. First, the husbandry procedures involved many of the same kinds of activities (bringing in equipment, checking each of the pens). Second, several blank trials were delivered before the start of experiments to allow the chickens to habituate. All procedures, including triggering the pneumatic system, were exactly the same during blank and experimental trials; only delivery of the playback differed. Data from the blank trials were used as a "baseline" or point of comparison that could be used to determine the effect of the overflights even if subtle responses to the experimental procedures were later discovered.

In the previous study by Bradley *et al.* small groups of turkey poults were used (1-4 groups of 25-200 poults), an approach that proved to be inadequate given inconsistent among-group variability in responses. Although domestic turkeys are not a good model for the domestic chicken in some ways, both species exhibit strong flocking behaviors; this meant that flock-specific responses were expected to be extremely important in chickens as well. For the purposes of these experiments, the sampling unit was taken to be the flock, even though some effort was made to collect data on individual chickens. To insure an adequate sample of flocks, small groups were kept in separate pens. Groups of 20-40 birds appeared to be large enough to obtain flock responses. Flocks responded as a unit, and were often relatively indifferent to the responses of neighboring flocks, even when they could still hear and see one another. A total of 14-16 pens per section were examined. This sample size was expected to be adequate to distinguish responses among groups based on the turkey study.

In the previous study of turkey poults (Bradley et al. 1990), birds piled and crowded during the first 1-2 exposures to overflight noise, but habituated quickly thereafter. After habituation, the only potential effects uncovered were a mild increase in activity and possibly an increase in irritability (more incidents of cannibalistic picking were found in exposed groups). During these experiments all behaviors likely to correlate with exposure, including abrupt movements (running, walking, getting up, alerting), maintenance behaviors (eating, drinking, preening), and agonistic behaviors (attacking or fleeing), were collected. One pen in the high exposure section was monitored by an observer during experiments to insure that observers had an immediate estimate of response.

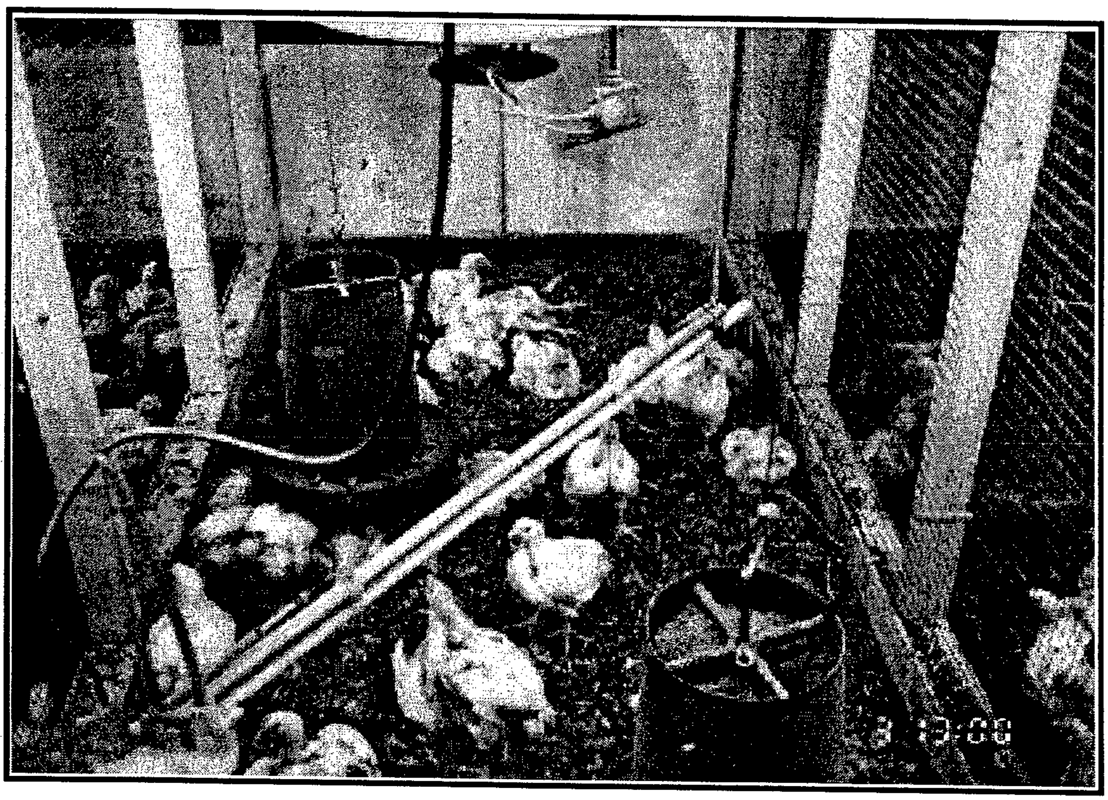

Figure 3. Photograph of 3-week old broiler flock in a test pen. The pens were equipped with a heater (far end top), two feeders, and an automated waterer (bar angled across center of pen). Pens were separated by a hardware-cloth barricade. Broilers could see and hear individuals in adjoining pens.

Table II. Playback experiments conducted on 8-week old broilers.

Date	Time Stimulus Started	Stimulus	Interval (min)
1/18/97	-	Baseline	-
1/19/97	-	Baseline	-
1/20/97	17:20:00	Blank	-
	17:40:14	1	20
	18:10:09	2	30
	20:20:31	3	130
	21:20:14	4	60
	00:00:09	5	160
1/21/97	12:45:20	3	-
	17:00:13	4	255
	17:10:07	5	10
	19:10:05	6	120
	19:40:14	1	30
	20:40:12	2	60
1/23/97*	10:10:17	3	-
	10:40:05	6	30
	10:50:11	1	10
	14:50:00	Blank	240
	15:50:11	2	60
	17:50:11	5	120
1/24/97	14:20:07	1	-
	15:20:00	Blank	60
	17:20:09	2	120
	17:50:21	3	30
	21:50:12	4	240
	22:08:18	5	20
1/25/97	10:20:17	3	-
	14:20:14	4	240
	15:20:18	5	60
	15:50:08	6	30
	16:00:00	Blank	10
	18:00:08	1	120
1/26/97	12:12:00	Blank	-
	12:25:00	Blank	10
	12:35:00	Blank	10
	13:05:00	Blank	30
	13:35:00	Blank	30
	13:45:00	Blank	10

*No experiments 1/22 due to equipment failure.

Table III. Playback experiments conducted on 3-week old broilers.

Date	Time Stimulus Started	Stimulus	Interval (min)
3/28/97	-	Baseline	-
3/29/97	-	Baseline	-
3/30/97	14:30:00	Blank	-
	15:30:00	Blank	60
	17:30:00	Blank	120
	18:00:00	Blank	30
3/31/97	10:40:25	3	-
	12:40:23	4	120
	13:40:32	1	60
	14:10:29	2	30
4/1/97	13:00:28	1	-
	16:40:08	2	220
	17:10:08	3	30
	18:10:04	4	60
4/2/97	17:00:05	3	-
	17:30:05	2	30
	19:30:08	4	120
	20:30:06	1	60
4/3/97	10:55:04	1	-
	12:55:03	4	120
	13:55:04	2	60
	14:25:06	3	30

Responses of flocks in other pens were collected from videotape and series of photographs from the still cameras.

During trials with turkeys, some individuals became irritable after exposure, resulting in an increase in the incidence of picking. During broiler experiments, an observer counted birds with evidence of picking twice daily (once before and once after the day's trials). Birds with blood on the comb or chest were counted. In addition, video and still images were checked for evidence of agonistic encounters.

Even if birds responded little to simulated overflights, it was possible that the exposures could alter productivity based on claims levied against the USAF (Milligan *et al.* 1983). To test this possibility, measures of productivity and health were collected in all three barn experiments, including food and water intake, bird weight, and egg production (laying hens). These measurements were being made regularly in the barns for other purposes and were familiar experiences for the birds.

PLAYBACK EXPERIMENTS WITH 8-WEEK OLD BROILERS

Eight-week old broilers were tested in mixed-sex flocks. A total of 1884 broilers were divided among the 72 pens in the experimental poultry house, averaging 26 birds per pen at the start of the experiment. All broilers became available as at 8 weeks of age. At that time, the birds were fully grown and were beginning to display physical characteristics and behaviors of adult males and females. Flocks had been fed a variety of diets throughout growth as part of the study that preceded these experiments.

Broilers were exposed to 4-6 different simulated jet overflights per day (Table II). Behaviors were measured as described above. Food consumption was measured every other day by weighing food containers before and after provisioning. Water consumption was measured by counting graduations in the container that supplied automated waterers. Weight was measured before and after the experiments by flock - birds in each pen were counted and weighed together in a big crate. The weight per unit bird was used as an outcome measure; in addition, weights were used to calculate average feed and water intake by weight for the pen. Dead birds were counted and removed once daily.

Five individuals per flock were marked with Malachite Green stain (a combination of spots on left wing, right wing, neck, and tail) for individual identification from overhead photographs. They

Table IV. Playback experiments conducted on laying hens.

Date	Time Stimulus Started	Stimulus	Interval (min)
5/31/97	-	Baseline	-
6/1/97	-	Baseline	-
6/2/97	11:15	Blank	-
	15:45	Blank	270
6/3/97	16:00	1	-
	18:00	2	120
	19:00	3	60
	19:45	4	45
6/4/97	12:15	3	-
	17:15	4	300
	18:15	2	60
	18:45	1	30
6/5/97	11:05	3	-
	13:00	2	115
	14:00	4	60
	14:30	1	30
6/6/97	10:00	1	-
	12:00	4	120
	13:15	2	75
	13:45	3	30
6/7/97	13:00	3	-
	15:00	4	120
	16:00	1	60
	16:30	2	30
6/8/97	15:30	Blank	-
	17:30	Blank	120
	18:30	Blank	60
	19:00	Blank	30
6/9/97	11:00	Blank	-
	13:00	Blank	120
	14:00	Blank	60
	14:30	Blank	30
6/10/97	10:00	Blank	-
	12:00	Blank	120
	13:00	Blank	60
	13:30	Blank	30

Table IV. Playback experiments conducted on laying hens, cont'd.

Date	Time Stimulus Started	Stimulus	Interval (min)
6/11/97	13:00	Blank	-
	15:00	Blank	120
	16:00	Blank	60
	16:30	Blank	30
6/12/97	15:30	1	-
	17:30	2	120
	18:30	3	60
	19:00	4	30
6/13/97	10:30	3	-
	12:30	2	120
	13:30	4	60
	14:00	1	30
6/14/97	10:10	1	-
	10:40	4	30
	13:25	2	105
	14:25	3	60
6/15/97	15:30	3	-
	17:30	4	120
	18:30	1	60
	19:00	2	30
6/16/97	09:50	1	-
	11:50	2	120
	12:50	3	60
	13:20	4	30

were sexed by the degree of development of the comb and wattles when possible.

Flock behaviors were monitored for 2 days (baseline measurements) before the start of trials. Six days of trials followed, with 6 experiments per day, for a total of 36 experiments; 26 of these were playback trials and 10 were blank trials. Each playback stimulus was delivered 4-6 times (Table II).

After the experiments were complete, a sample of 140 birds was slaughtered. Weight of the carcass and notations of bruising or other possible carcass quality problems were recorded by an experienced slaughterhouse supervisor.

PLAYBACK EXPERIMENTS WITH 3-WEEK OLD BROILERS

Broilers for this experiment were obtained as hatchlings (< 5 days of age) for acclimation to the experimental housing, but were not exposed until they were 3 weeks of age. This age was chosen because younger birds do not have fully-developed hearing (Gray and Rubel 1985) or fully-

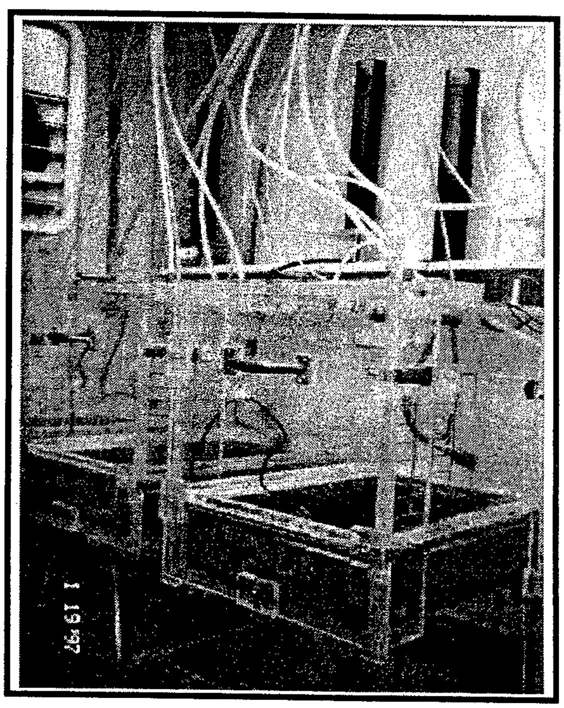

Figure 4. Photograph of metabolic chamber.

developed startle responses, and rarely harm themselves in panics (Von Rhein 1983).

72 pens of 60 3-week-old broilers were tested (a total of 4388 birds). Behavior, food and water consumption, and changes in body weight were measured as described above. Five broilers in each pen were marked with Malachite Green dye for individual identification in photographs and on video.

Broilers were exposed to 4 simulated jet overflights per day (stimuli 1-4, Table I). A total of 16 playback experiments (4 trials with each of 4 stimuli) and 4 blank trials were completed (Table III). In addition, two days of baseline measurements were completed before the start of trials.

PLAYBACK EXPERIMENTS WITH LAYING HENS

The hens used in these experiments were Cobb-Vantress layers that had just passed the peak of laying output. They were made available at the end of an experiment to measure laying efficiency and bone density under different feed regimes (Teeter 1998 unpub report). Laying hens are productive for many years, but their peak of productivity occurs in the first 30-35 weeks of life (Figure 2). The hens used in these experiments were 48-52 weeks of age. Hens were therefore of known age, in good health, had established reproductive histories, and were laying at very predictable rates. These data could be used to estimate expected production throughout the playback experiments.

Table V. Design of stimuli used for physiological response measurements on 3-week old broilers. Six tapes with 7 stimuli were prepared from 26 different stimulus variants. The breakdown of these stimuli by feature (level, onset, and duration) is given.

Number of Stimuli	ASEL	High Onset	Low Onset	Short Duration	Long Duration
6	105	3	3	3	3
7	95	3	4	4	3
7	85	3	4	4	3
6	75	3	3	2	4

Hens were housed in a 60-pen house. Fifty-six pens with 30-32 hens were used in the experiments. They were exposed to 5 days of simulated overflights (4 exposures/day with 4 stimuli), a 4-day hiatus in which 4 blank trials were delivered daily, and 4 more days of simulated overflights (Table IV). The wait between the two series of exposures was designed to permit laying patterns to recover from any effects of the initial exposure series.

Hen behavior, weight, food consumption, egg production, and egg quality were measured in addition to behaviors. Eggs were laid in nesting boxes designed to allow easy removal of eggs, which were counted, weighed, and candled daily by flock. Counts of eggs with double yolks and other abnormalities were tallied. Mortalities were very low, too low to provide a useful outcome measure.

PHYSIOLOGICAL MEASUREMENTS

A small sample of 3-week-old broilers was taken from the same population as above. An experienced veterinarian (Dr. VanHooser, D.V.M.) implanted a MiniMitter PhysioTel CTA series heart-rate transmitter (CTA-F40) in the body cavity under surgical conditions.

A total of 6 broilers were fitted with the implanted heart rate monitors. After surgery, birds were placed in a plexiglass metabolic test chamber and permitted to recover for several days. The test chambers measured 25 x 30 x 15 in (Figure 4). The chambers were small enough to allow reasonably good reception from the transmitter for most positions the bird elected to adopt.

Surgical implantation of the heart rate monitor selected because birds tolerate surface-mounted electrodes poorly. During one short test, an effort was made to calibrate the MiniMitter monitor *in situ* by making simultaneous recordings with a SmartHeart monitoring system connected to surface-mounted electrodes. A total of 3.3 hr of simultaneous monitoring was completed, but the bird preened persistently until it succeeded in pulling off the surface electrodes. No usable data were collected.

Birds were exposed to a series of 14 experimental overflights per day at random intervals for 2 weeks. Overflights varied in absolute level, onset time, duration, and inter-stimulus interval (Table V). Time-lapse video and an LD 820 community noise monitor were used to monitor behavior and the corresponding exposure. Level was measured outside the chamber during experiments. Level inside the chamber was estimated by collecting a few measurements inside the chamber before the start of the trials with the chicken absent.

ANALYSIS OF DATA

In both the 72-pen and 60-pen barns, total of 14-16 pens per sound level category (section) were monitored. The number of birds rising and congregating was selected as an outcome variable *a priori* based on previous experience. However, as this proved to be an unusual response, other variables were examined as well during *post hoc* analysis.

The criteria for evaluating outcome measures was as follows:

For the dose-response model

1. Frequency distribution of the measure must be approximate a normal distribution ($\alpha = 0.001$); transformation to achieve this goal is allowable
2. Inter-pen variance must be low
3. Variance of the measure must be as homogeneous as possible
4. The measure must scale directly with dosage in the range from 65-120 dB ASEL

Latencies were likely to make good outcome measures for a dose-response model in pens monitored using video because they could be observed at all sound levels and because they have varied directly with stimulus level in previous experiments.

In the previous study of turkey poults (Bradley *et al.* 1990), response latencies were compared among treatment conditions (*e.g.*, exposed *vs.* unexposed) using parametric multiway ANOVA after appropriate transformations. The same approach will be adopted here. The relationship between measures of exposure and responses will be determined using non-linear regression. Based on previous experiments, a logistic growth model should fit the data. However, the number of replicates per condition will be small, making it possible that simpler models (e.g., linear growth) will fit equally well. The ideal solution would be to conduct more trials to insure sufficient statistical power to distinguish among the possible models clearly. However, such effort is outside the scope of this program. For this reason, the final results will include both the best fit model and the logistic growth model.

Picking will be compared among conditions using the Chi-square test.

The results of the study will be integrated into the USAF dose-response model for domestic fowl. This will include developing an empirical model for the habituation process and a dose-response curve analogous to the Schultz curve for humans. It will also attempt to allow for the effect of group size, interval between exposures, and stimulus parameters (based on the results of the metabolic rate experiments). The results will be provided in a form suitable for entry into the USAF model.

PLAYBACK EXPERIMENTS WITH 8-WEEK OLD BROILERS

Initial jet simulations at the highest levels stimulated 8-week old broilers to stand, aggregate, and search for the source of the sound visually. Although a few ran briefly, none of the groups piled and crowded. After the experiments, HSWRI and OSU staff suggested possible explanations for failure to panic: broilers (1) were past the age of maximal responsiveness, (2) were accustomed to human-made disturbances, or (3) were in groups that were below some 'critical mass' needed to produce panic responses. Of these, the first was deemed the most likely, so a second series of experiments with smaller birds was planned (3-week old broiler experiments).

With repeated exposure, broilers ceased to stand or aggregate. By the last day of experiments, broilers looked up or simply remained still during the simulations. However, they exhibited a highly stereotypical sequence of events after the exposures, regardless of their initial reaction (Figure 5). At <2 min post-exposure, they relaxed. At 2-4 min, they preened more, drank more and fed more than during the pre-stimulus period. From 2-8 minutes, they were more active and engaged in more aggressive encounters. After this, they returned to baseline activity levels.

This sequence of events was apparently an innate response to disturbance. Entry of humans into the pen to care for the broilers and other husbandry activities produced a similar sequence of behaviors.

During the 8-week-old broiler experiments, incidents of picking were quantified by collecting counts of birds with comb wounds or blood on their feathers twice per day (14 observations/pen were collected in the baseline and experimental period). These incidents were seen most often in the section of the barn where human activity was greatest (4), but also correlated with noise exposure. In the low exposure section (3) of the barn, 48 birds were seen with blood spots; 62 were seen in the moderate exposure section (2) and 98 were seen in the high exposure section (1). Human activity was highest in section 4, which also experienced elevated ambient noise levels from the ventilation system; 91 birds were observed with blood marks in this section (Figure 6). Human activity was comparable in sections 2 and 3. One bird died due to picking at the exposed end of the barn (section 1). Although human activity confounded the results of these measurements, making it impossible to demonstrate statistically that picking was stimulated by noise alone, the data did lead to an important conclusion – picking results directly from arousal and congregation brought on by disturbance. Congregation brings birds into close proximity, after which rivals initiate

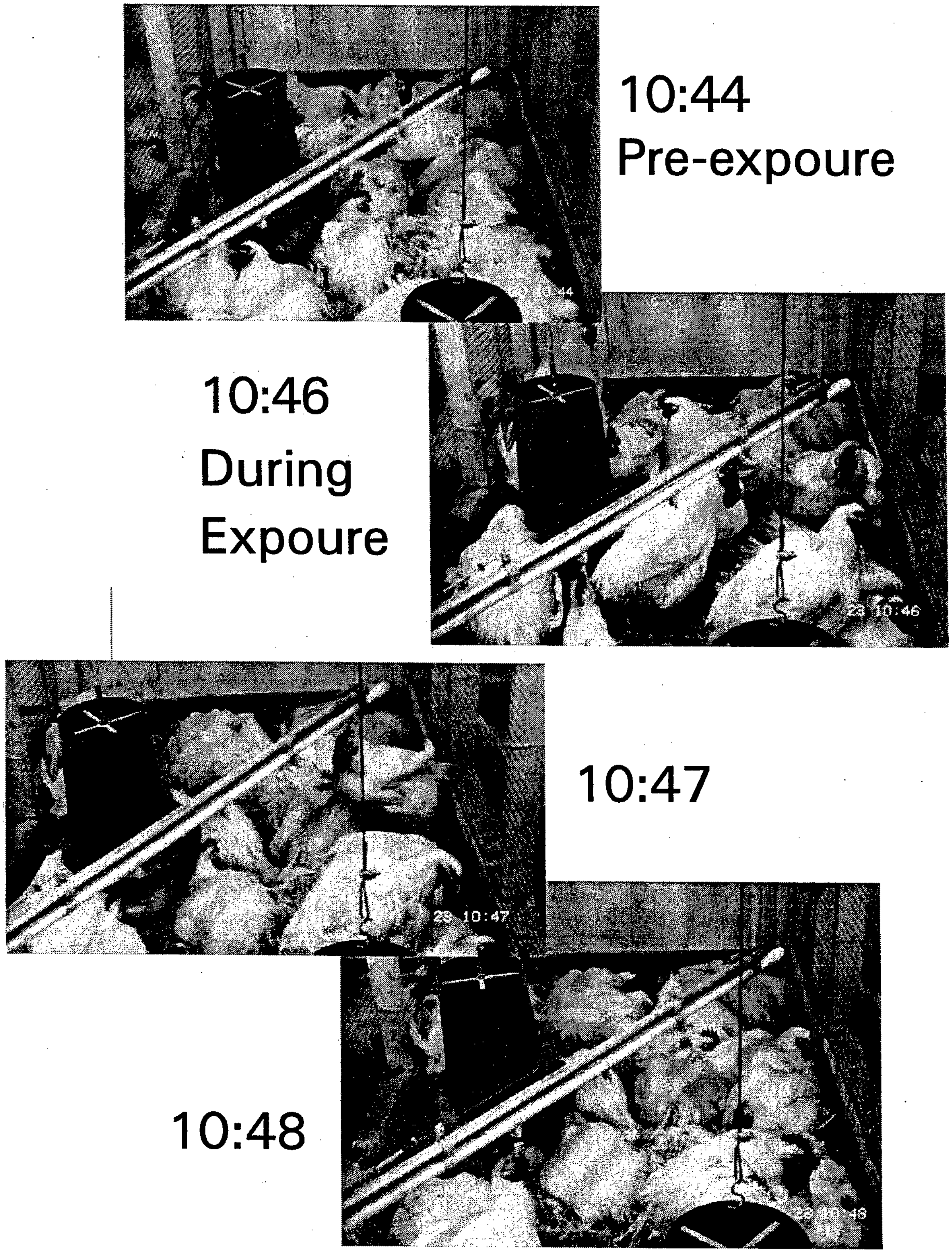

Figure 5. Responses of 8-week old broilers to playback stimulus. The photographs show a concentration of broilers in the vicinity of food containers and an automated waterer. Initially, most broilers were inactive (10:44). Immediately after the overflight (10:46), many broilers rose, oriented, ate and drank. By 1 min post-exposure (10:47), they had begun to preen and rest again. By 10:48, they were quiescent again.

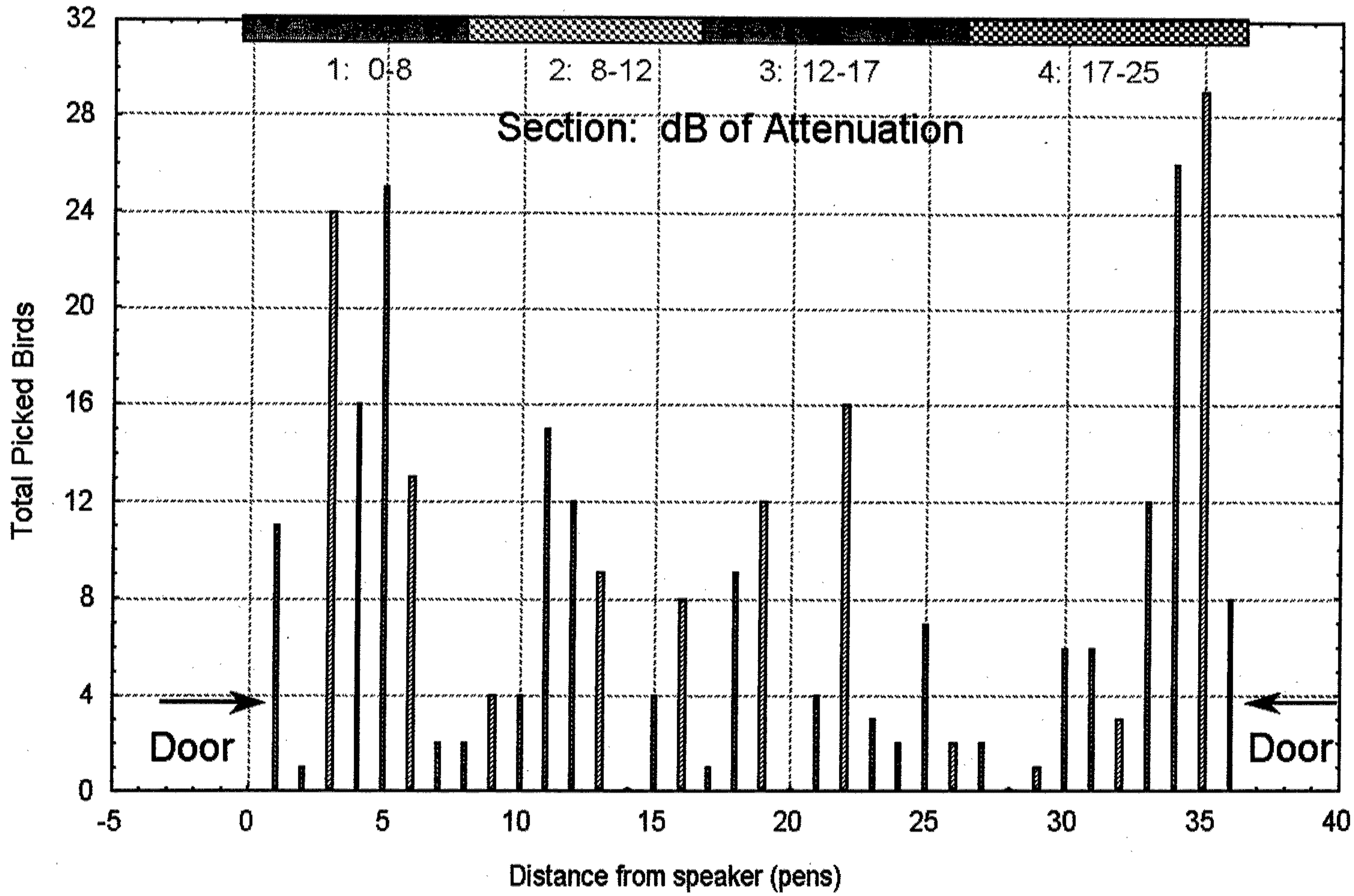

Figure 6. Incidence of picking in the four segments of the 72-pen poultry house during experiments with 8-week old broilers. The total number of birds counted during the experiments is shown by pen. Playback loudspeakers were located near pen 0; husbandry activities took place throughout the barn. Humans usually entered and left through a door near pen 36.

aggressive interactions.

No detectable differences in growth rate or mortality were observed among exposure conditions in 8-week-old broilers. Mortality was highest in the 8-week old broilers - an average of 14% of the birds died of various causes during the course of the test. The subject birds were just past the commercial age of slaughter; at this age, growth was very rapid and crowding became an important factor. No significant differences were found among the section of the barn in mortality, however (ANOVA, $p >> 0.05$; the highest mortality was found in the low-exposure section).

Broilers grew by an average of 0.58 lb during the experiments, but there were no significant differences among barn sections in weight gain (ANOVA, $p >> 0.05$), despite significant differences in initial and final weight.

PLAYBACK EXPERIMENTS WITH 3-WEEK OLD BROILERS

After exposure, 3-week old broilers exhibited the same sequence of behaviors as 8-week old broilers, excepting that they did not become aggressive when in close proximity. Three-week old broilers were rarely aggressive to one another and did not pick regardless of the disturbance source. Initial jet simulations at the highest levels stimulated young broilers to stand, aggregate, and search for the source of the sound visually. Although a few ran briefly, none of the groups piled and crowded. Therefore, age was probably not a crucial factor in determining piling and crowding behavior.

With repeated exposure, broilers ceased to stand or aggregate. By the last day of experiments, broilers looked up or simply remained still during simulations. However, they exhibited a highly stereotypical sequence of events after the exposures, regardless of their initial reaction. At 1-2 min post-exposure, they relaxed. At 2-4 min, they preened more, drank more and fed more than during the pre-stimulus period. From 4-8 minutes, they were more active than before the overflight, returning to baseline activity levels at the end of this period.

No detectable differences in growth rate (Figure 7) or mortality were observed among exposure conditions in 3-week-old broilers. Mortalities were generally very low in this cohort of broilers.

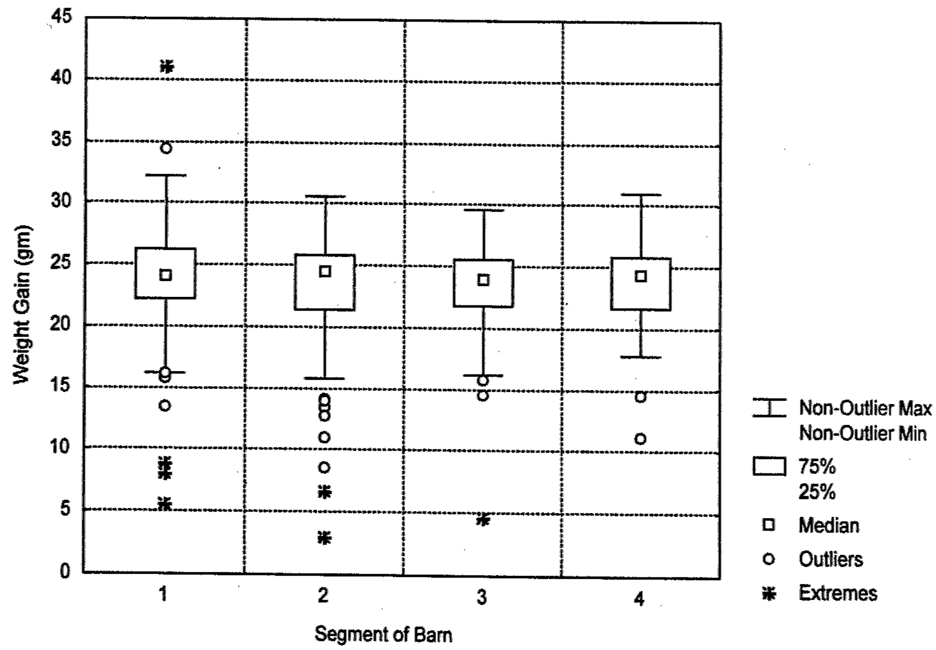

Figure 7. Weight gain of 3-week old broilers by segment. No significant differences were found by segment. The highest weight gains were observed in the segment with the highest exposure.

PLAYBACK EXPERIMENTS WITH LAYING HENS

Hens followed much the same sequence of behaviors as broilers during and after exposure, but were less active. Hens occasionally stood, but never ran, even during initial exposures. The strongest response was usually alerting, after which increases in foraging, drinking, and aggression were so rare that they could not be detected statistically. No pecking incidents were recorded.

As was found in experiments with turkey poults (Bradley *et al.* 1990), hens habituated rapidly to aircraft simulations. Figure 8 shows the results of detailed observations made in one pen by an observer during experiments. The observer measured latency to relax, the time required for all hens in the flock to cease active orienting (alert with head high; looking around) and return to other activities. Initial exposures aroused extensive orienting, with hens looking for several minutes before relaxing. Within 3 trials, hens looked only briefly. Interestingly, they did not become completely unresponsive - when their behavior was compared between blank and playback experiments, a small but significant difference was found. Hens exposed only to the clicking of the pneumatic system did not look around or did so only briefly (15 s or less). Hens exposed to overflights looked around for periods of 15-30 s. As was the case for the broilers, many hens were

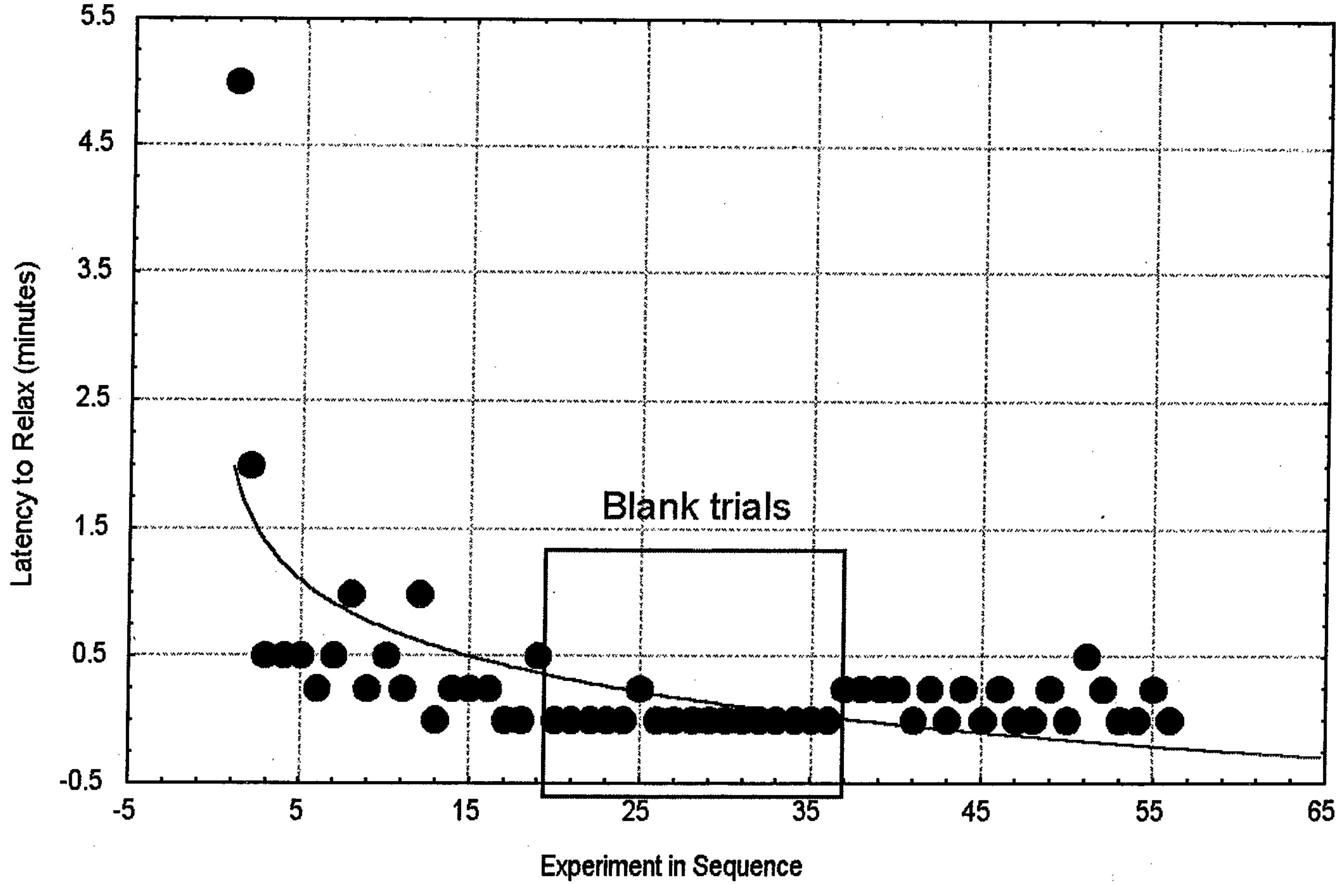

Figure 8. Habituation of hens in a single pen during playback trials. Latency to relax is the time required for all hens to relax from the alert or looking postures after exposure. Hens were exposed to 5 days of 4 playback experiments, then 4 days of blank trials, then another 4 days of exposures. Responses during blank trials are shown in the box.

motivated to orient on the source of the sound, but they often looked around in other directions as well (particularly toward the entryway at the other end of the barn). The incidence of calling changed during this period, with hens becoming silent briefly during and immediately after simulations and somewhat more vocal afterwards.

Hen behavior varied with simulated aircraft type. More than 90% of the hens in a flock alerted in response to the high-onset F16 simulations, whereas only 50-70% alerted in response to others (Figure 9), even though the durations were generally longer.

Hens had a known laying history before the start of experiments and were in the linear, declining phase of productivity. No difference was found between observed and expected laying rate during exposure days, either when compared with the previous history for the flock or the 4-day hiatus between exposure periods. Figure 10 and 11 show the decline in productivity of laying hens in all four sections, starting from 260 days of age (~37 weeks). This decline was linear, with variation on the order of 0.2 eggs/hen/day. There was no detectable change in the trend by section during exposure experiments (Figure 9).

The number of eggs/hen and weight of eggs/hen did not vary by section of the barn (Figure 12, 13). However, the largest eggs were laid in section 4 and the smallest in section 1. The numbers of

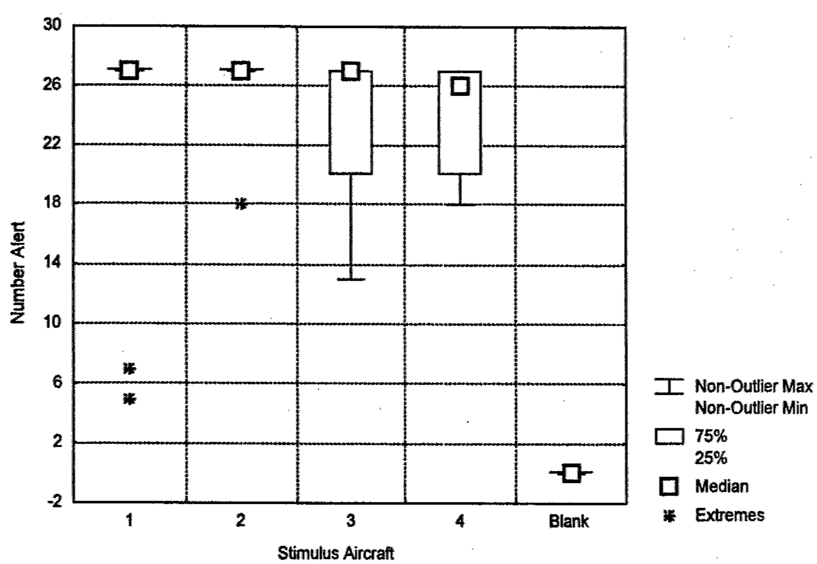

Figure 9 .Number of hens alert by stimulus type. Most of the hens in each pen alerted immediately after exposure to stimuli 1 and 2 (rapid onset). As few as half alerted after exposure to stimuli 3 and 4 (slow onset).

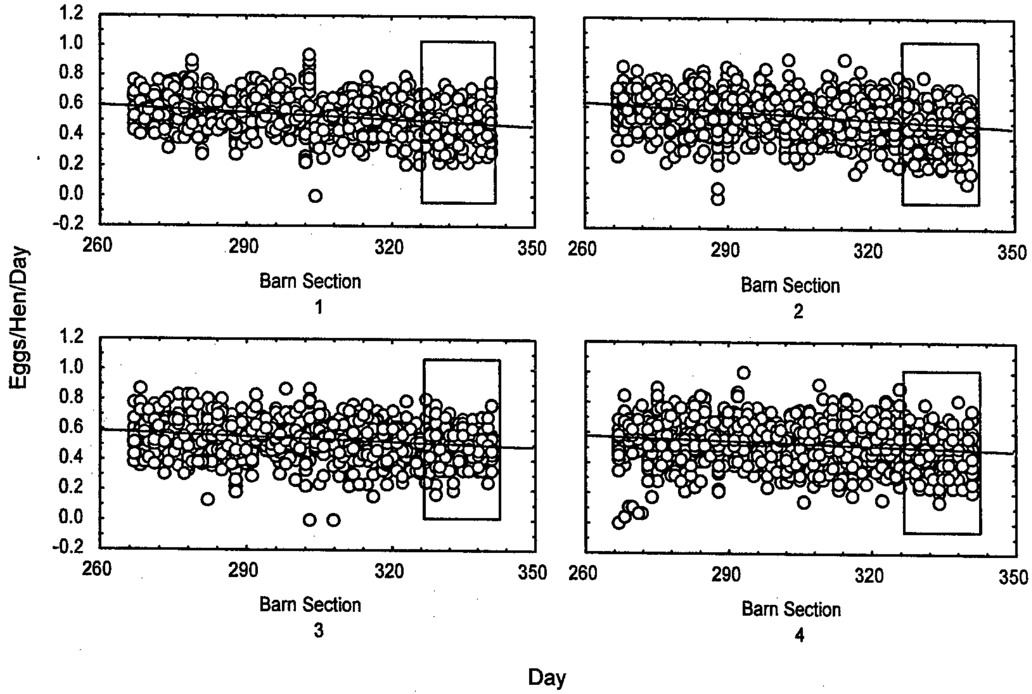

Figure 10. Eggs/hen/day for all pens by barn section (section 1: highest exposure; section 4: lowest exposure). Laying rates from age 37 weeks onward are shown; the slow decline in laying rate is normal. Laying rates during experimental exposures are indicated by box.

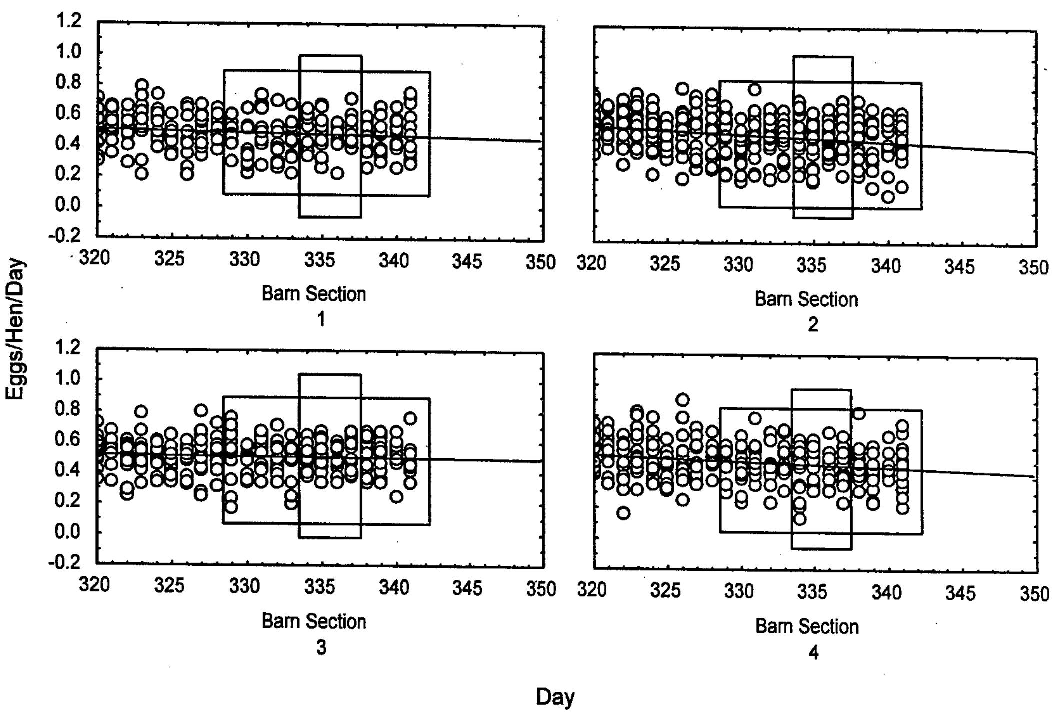

Figure 11. Eggs/hen/day for all pens by barn section (section 1: highest exposure; section 4: lowest exposure). Laying rates during experiments are shown. Laying rates during experimental exposures are indicated by wide box; laying rates during blank trials indicated by narrow box.

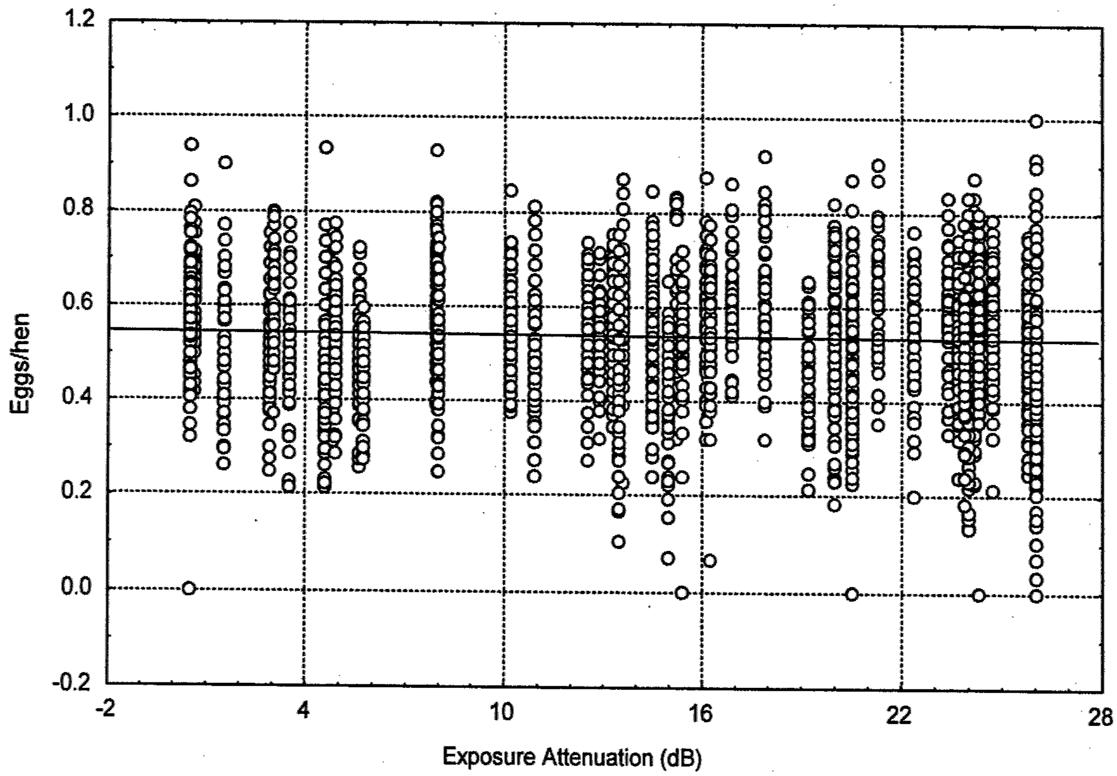

Figure 12. Eggs/hen/day plotted against attenuation for all flocks by segment.

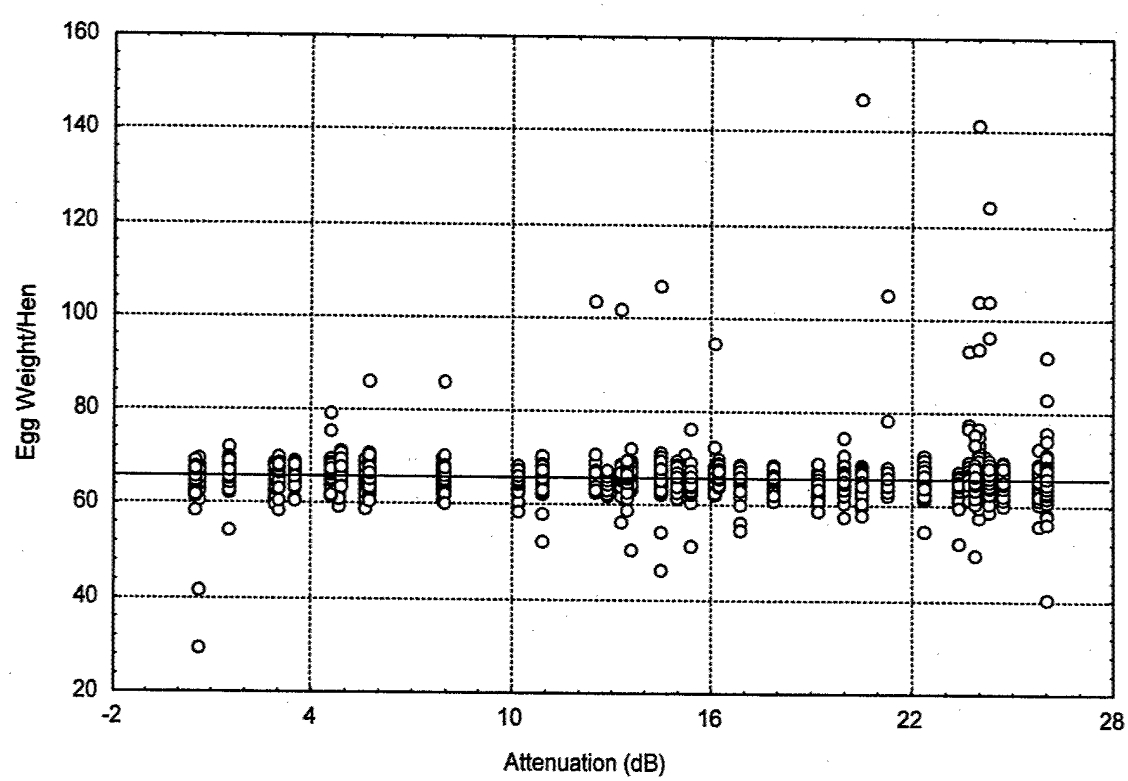

Figure 13. Weight of eggs/hen/day plotted against attenuation by segment.

unusually large and small eggs were too small to have economic impact (out of a total of 4200 counts, 19 counts yielded values > 95% confidence interval and 17 counts < 95% confidence interval).

Because a previous study (Kagan and Ellis 1974) had found blood spots in eggs of hens exposed to continuous noise, there was concern that exposure to aircraft noise might affect egg quality. On one day before exposure and one day after the first block of exposures, which would have been most likely to alarm hens, eggs were candled to determine whether they contained double yolks, blood spots or tissue spots. Double yolked eggs occurred at low incidence, generally < 0.08 eggs/hen. During the 40th week of life, many hens produced double yolked eggs (rate up to 0.23 eggs/hen). The reasons for this increase are unknown; the change was transitory and preceded overflight experiments. No change was observed during experiments (rates 0.02-0.03 eggs/hen). In the pre-exposure candling, 0.4% eggs/hen had blood spots and 0.5% had tissue spots (of 790 eggs). There was no significant difference post-exposure, although the magnitude of the incidence declined (0.1% had blood spots and 0.1% had tissue spots out of 740 eggs).

In summary, hens and broilers exhibited a highly-stereotypical sequence of behaviors after exposure, including alerting, becoming more active (increased walking, eating, and drinking), occasionally followed by agonistic (=aggressive) interactions as birds came into proximity in congregations or at the feeding and drinking stations. No piling and crowding incidents were observed in the small flocks that were tested (pens had up to 60 individuals). An increase in picking was observed in older (8-week-old) broilers that was not observed in younger broilers or hens. The number of birds encountered with marks of picking correlated with levels of exposure to disturbance; it is possible that simulations contributed to the disturbance in the barn, but effect of noise could not be distinguished from necessary human activity in the barn. While the degree of loss due to this cause was small (at most a few birds), the behavior can also affect carcass quality and bird welfare. The effect required repeated exposures.

Hens and broilers did not experience excess mortality or changes in weight as a result of exposure. Physiological measurements (heart rate and metabolic rate) and effects on feed and water consumption have not been analyzed as yet.

Egg production was not affected by simulated overflights, nor was egg quality.

Failure to pile and crowd remains unexplained. Aggregation, a precursor to this behavior, was observed during the first few exposures in the most heavily exposed flocks. During video and photographic analysis, aggregation (3 or more birds coming within 1 body length) was treated as an indicator of response for dose-response curve estimation. However, in experiments on turkey poults (Bradley *et al.* 1990) dangerous piling and crowding was aroused easily by simulated overflights. Therefore, it seems unlikely that susceptibility to panic in chickens is determined by age-class, sex, or small differences in genetic makeup (hens were from a different strain of the white leghorn than broilers). It seems most likely that chickens simply do not have a strong tendency to panic. In chickens, group size may also determine susceptibility, but large groups of the type found in commercial chicken operations were not tested.

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